

FIG. 1A

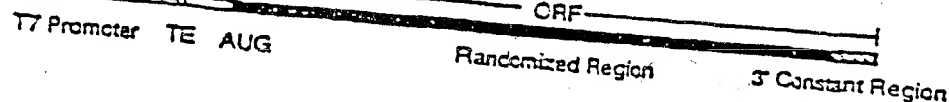


FIG. 1B

Puromycin-tethered oligo is ligated to mRNA (generated from above construct) in the presence of a splint and DNA ligase

note: for short ORFs, this whole template can be made synthetically

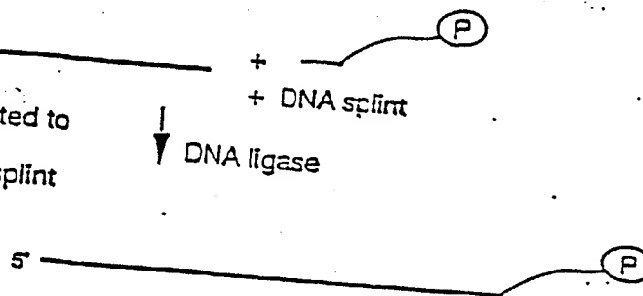
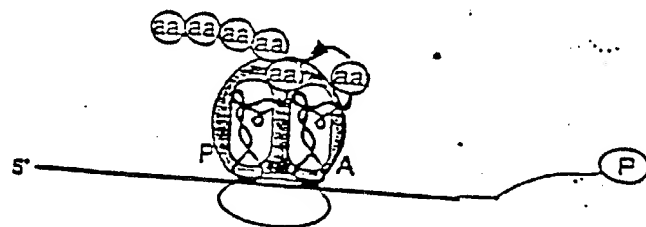
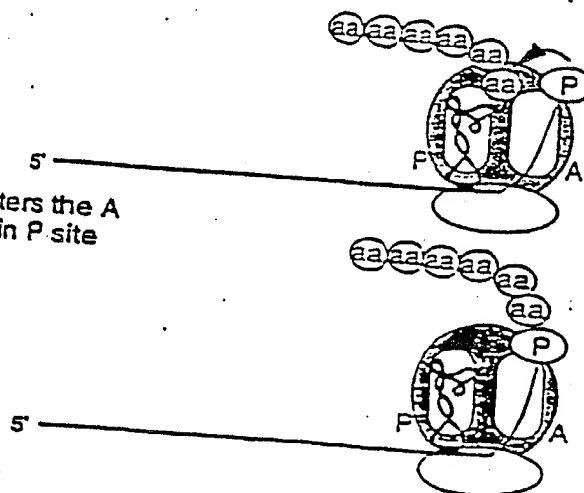


FIG. 1C

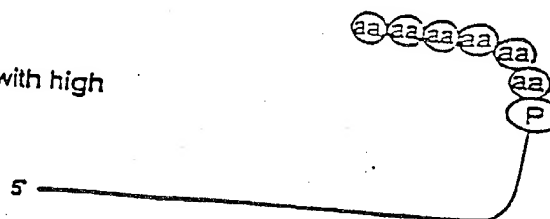
in vitro translation proceeds normally from the 5' to the 3' end of the mRNA



covalently linked puromycin enters the A site and attacks peptidyl tRNA in P site



Release of RNA-protein fusion with high salt wash of Ribosome



KEY

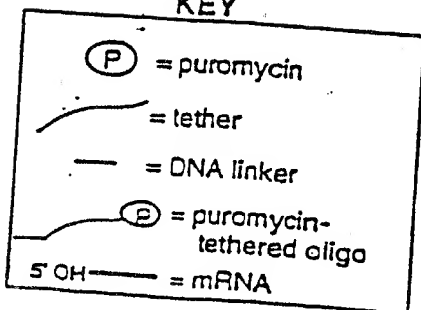


Figure 1 consists of 12 histograms arranged in a single row. Each histogram represents the distribution of the number of non-zero elements in the vector x for a specific value of n . The x-axis for all histograms is labeled 'x' and ranges from 0 to 120. The y-axis is labeled 'count' and ranges from 0 to 1000. The histograms are for $n = 10, 20, 30, 40, 50, 60, 70, 80, 90, 100, 110, 120$. As n increases, the distribution of non-zero elements shifts to the right, indicating that more elements in the vector x are non-zero for larger n . The peak count for each distribution decreases as n increases.

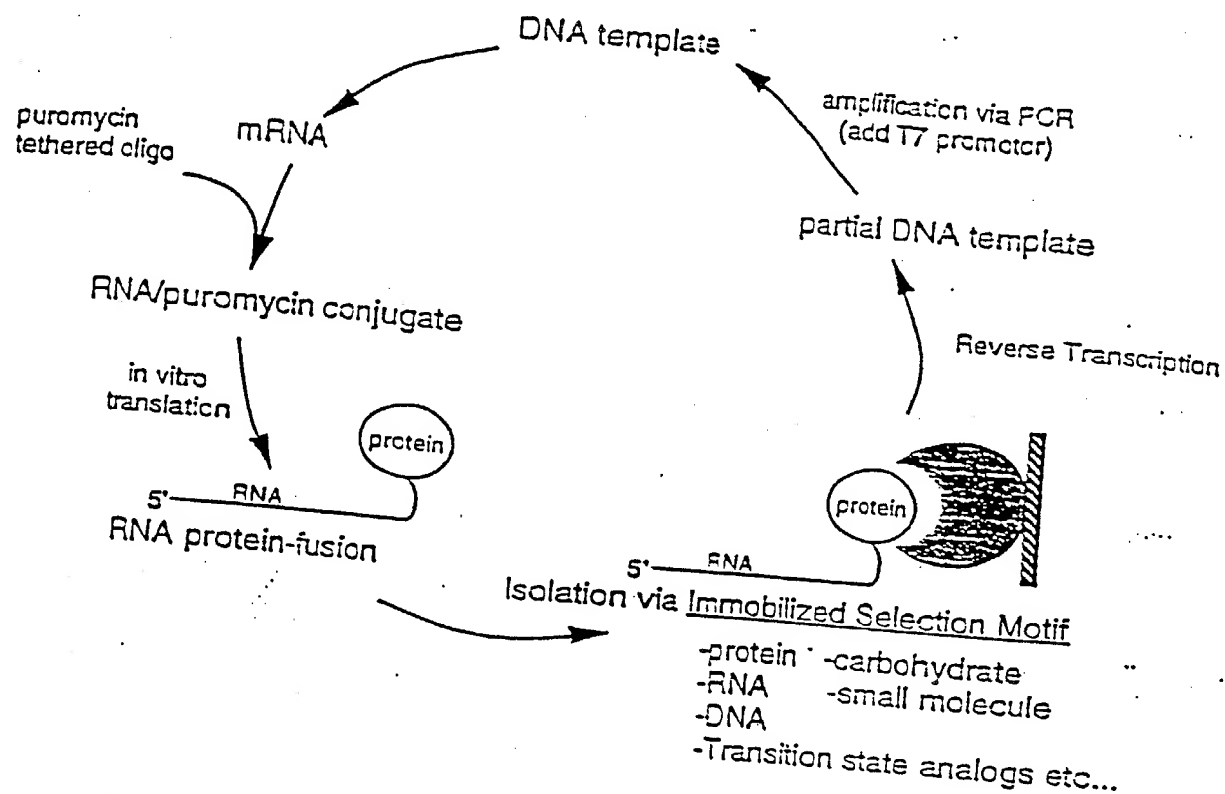
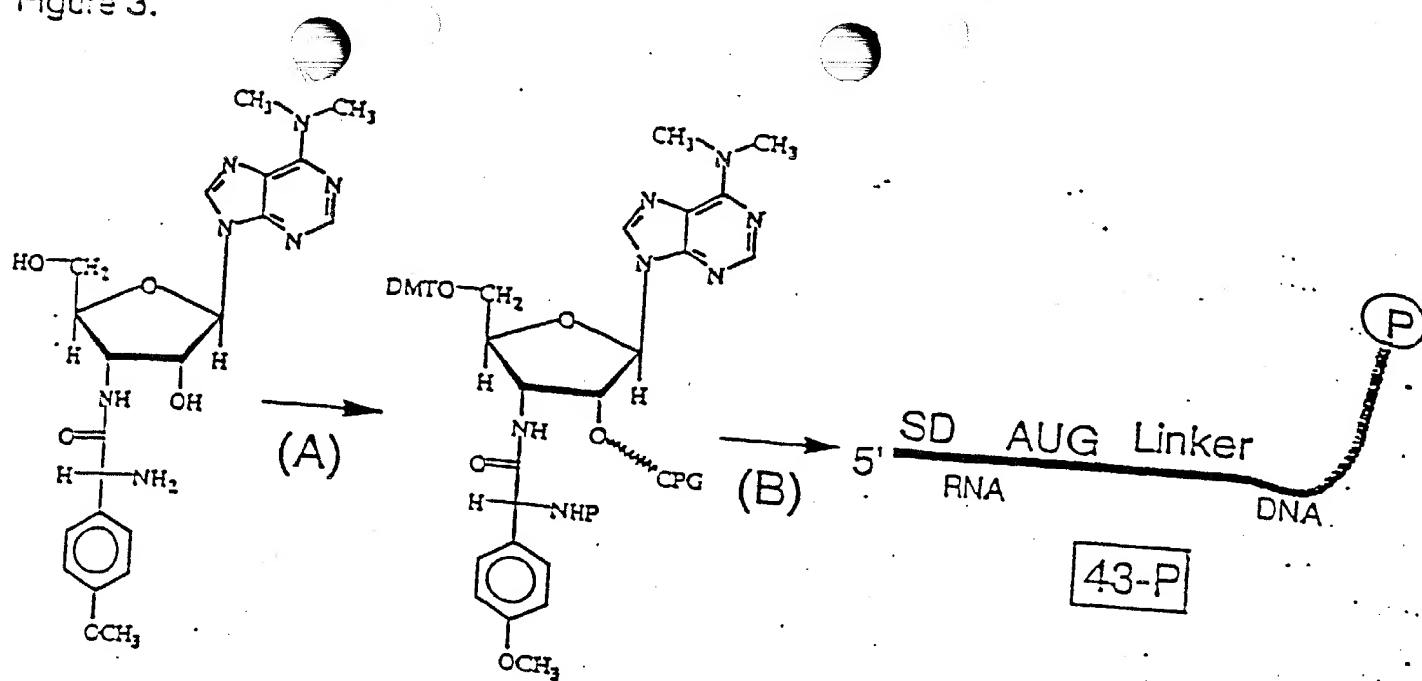
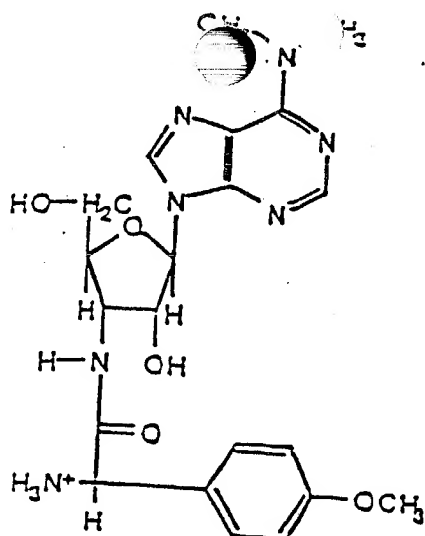


Figure 3.



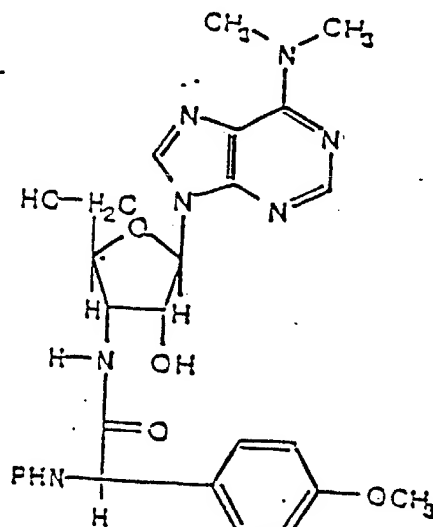
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FIG. 4



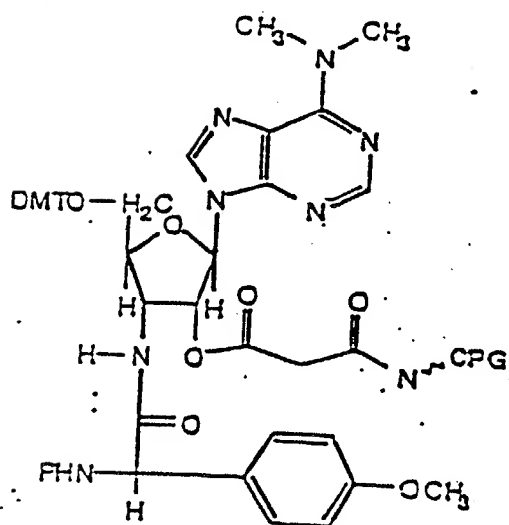
puromycin

protect amino group
with $(\text{CF}_3\text{CO})_2$



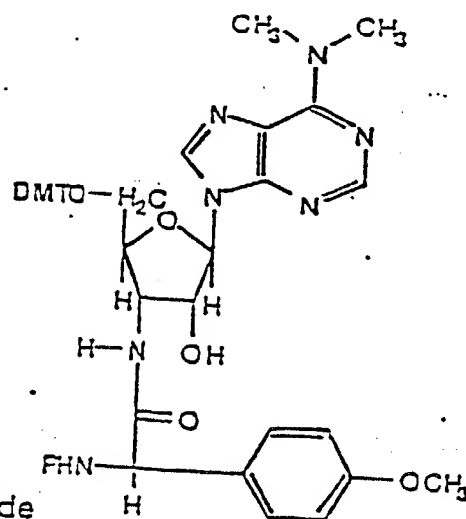
P = protecting group

protect 5'OH with
trimethyl chloride



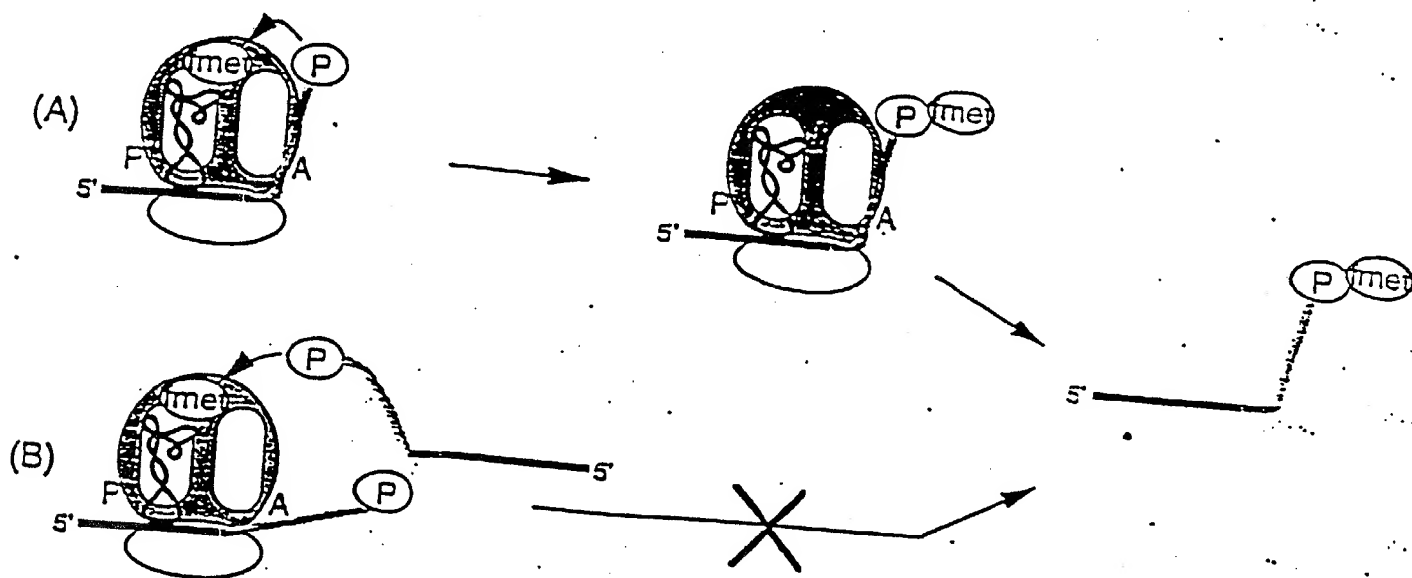
protected, CPG puromycin

link 2'OH to solid
support alkyl amine
CPG with
1) DCC/succinic anhydride
2) *p*-nitrophenol



Use as solid support in
automated DNA synthesizer
-deavage yields 3' puromycin
tethered oligonucleotide

FIG. 5



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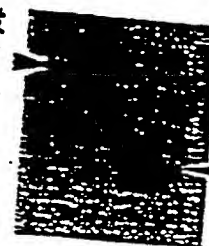
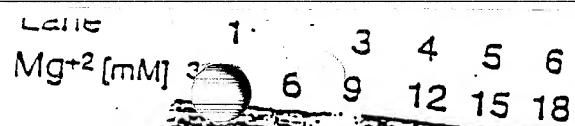


FIG. 6A

FIG. 6B

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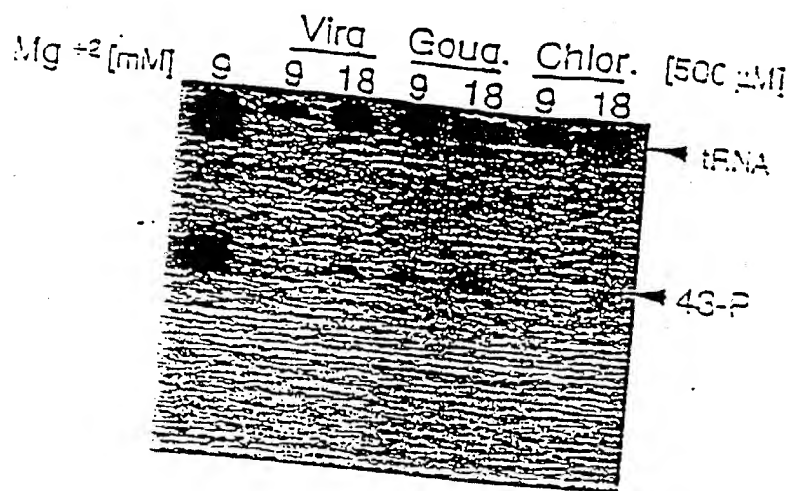


FIG. 6C

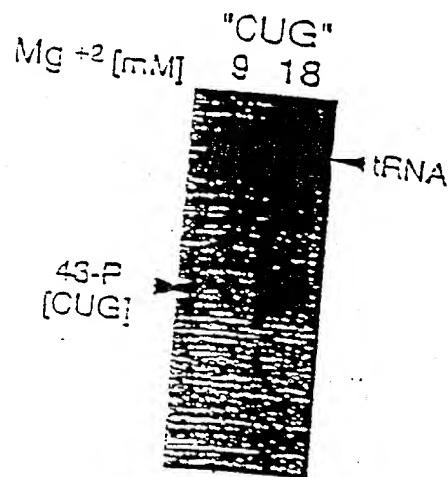


FIG. 6D

Linker (dA) _n	n=27		n=24		n=21		n=18	
Mg ²⁺ [mM]	9	18	9	18	9	18	9	18



FIG. 6E

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43-P +
25-P +

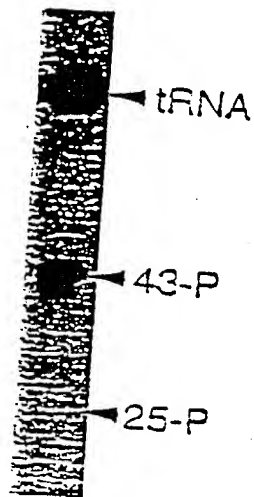


FIG. 6F

43-P + +
13-P - +

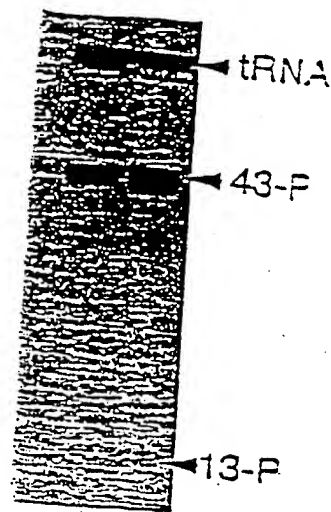


FIG. 6G

30-P - + + -
43-P + - + -

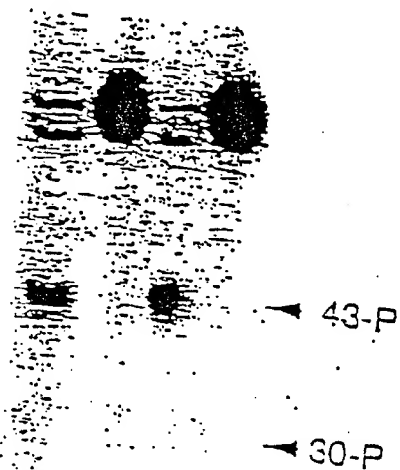


FIG. 6H

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FIG. 7A

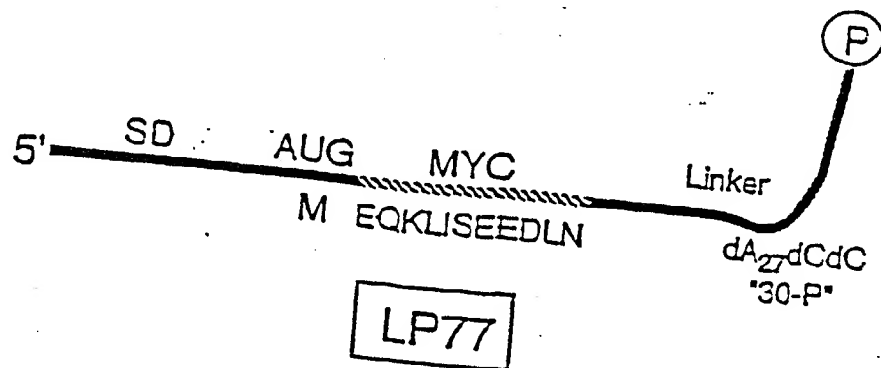


FIG. 7B

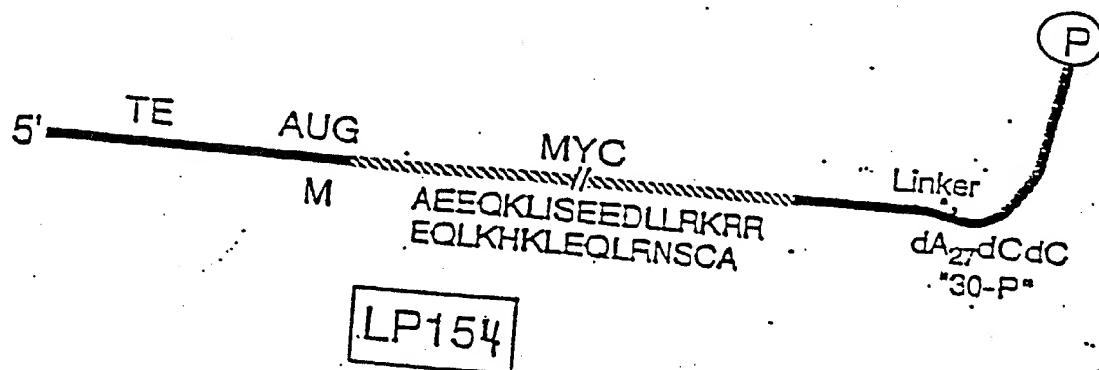
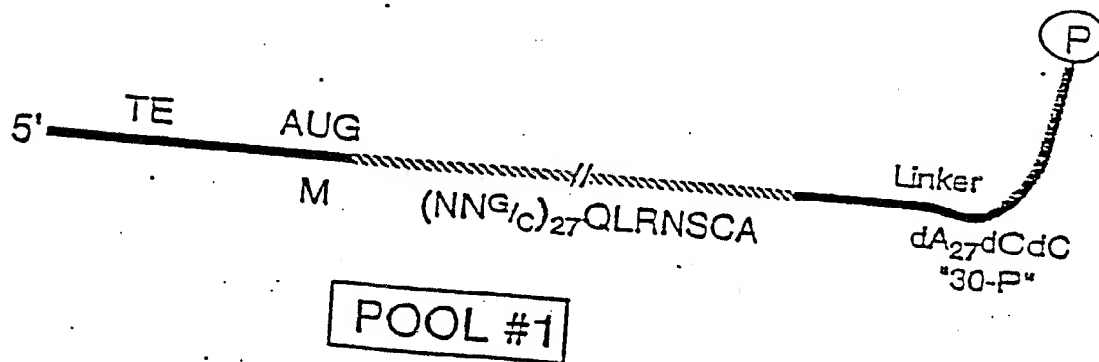


FIG. 7C



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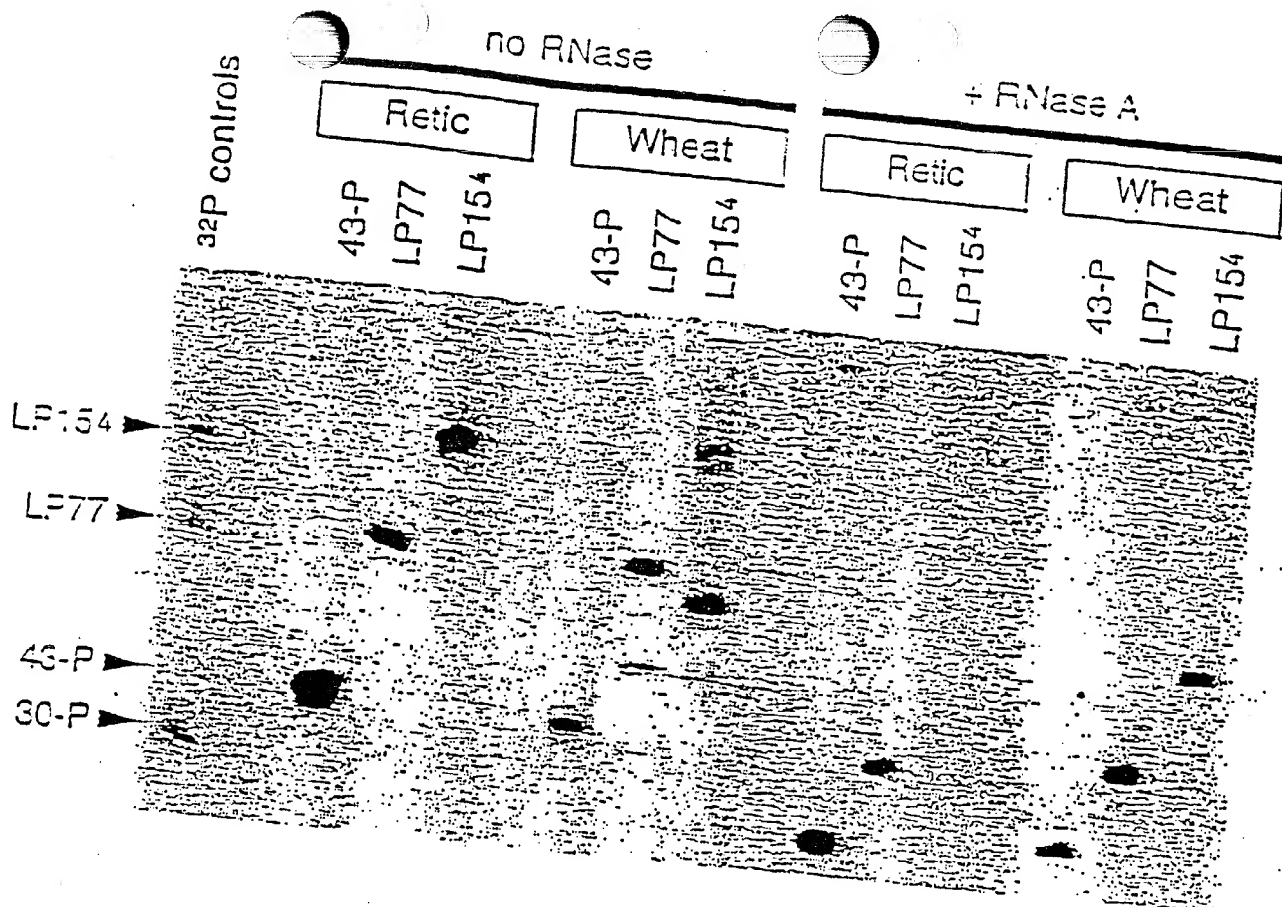
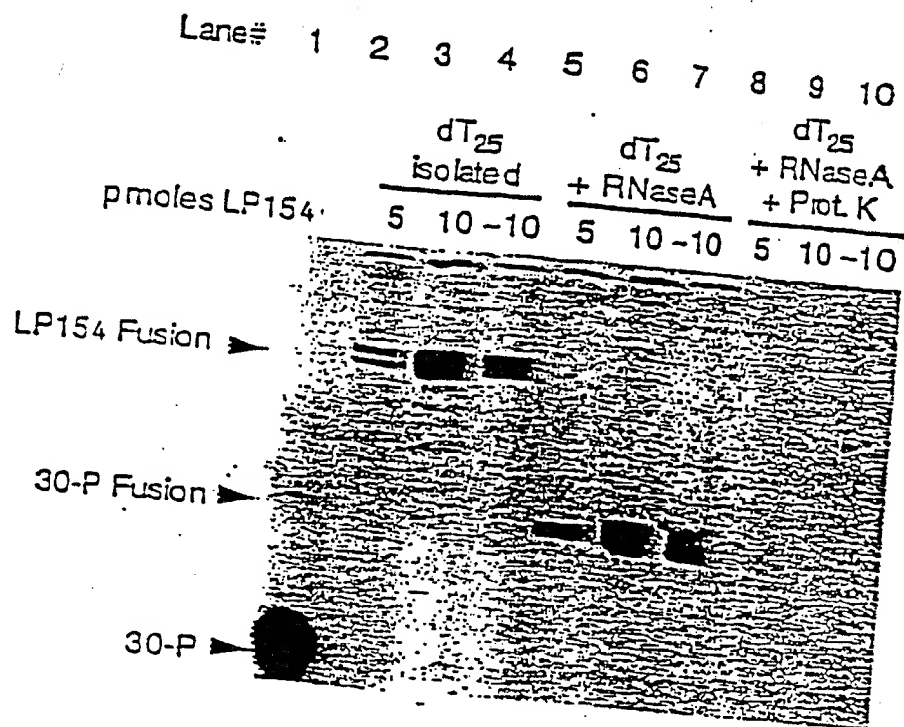


FIG. 8

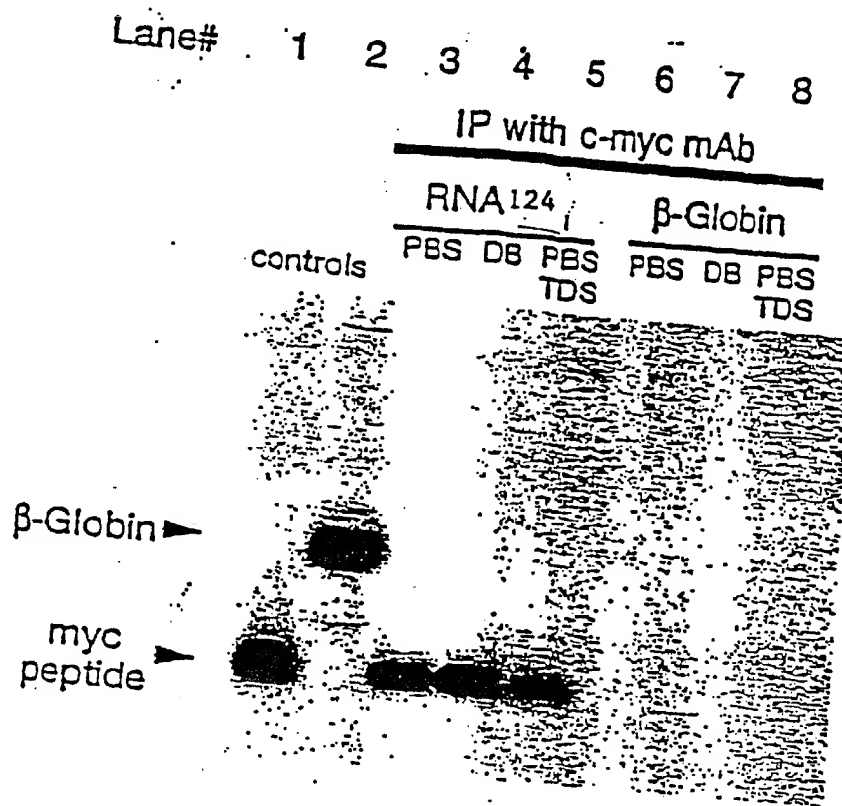
FIG. 9

Effects of RNase A and Proteinase K on fusion



0007625 160501

FIG. 10



087625-060901

FIG. 11

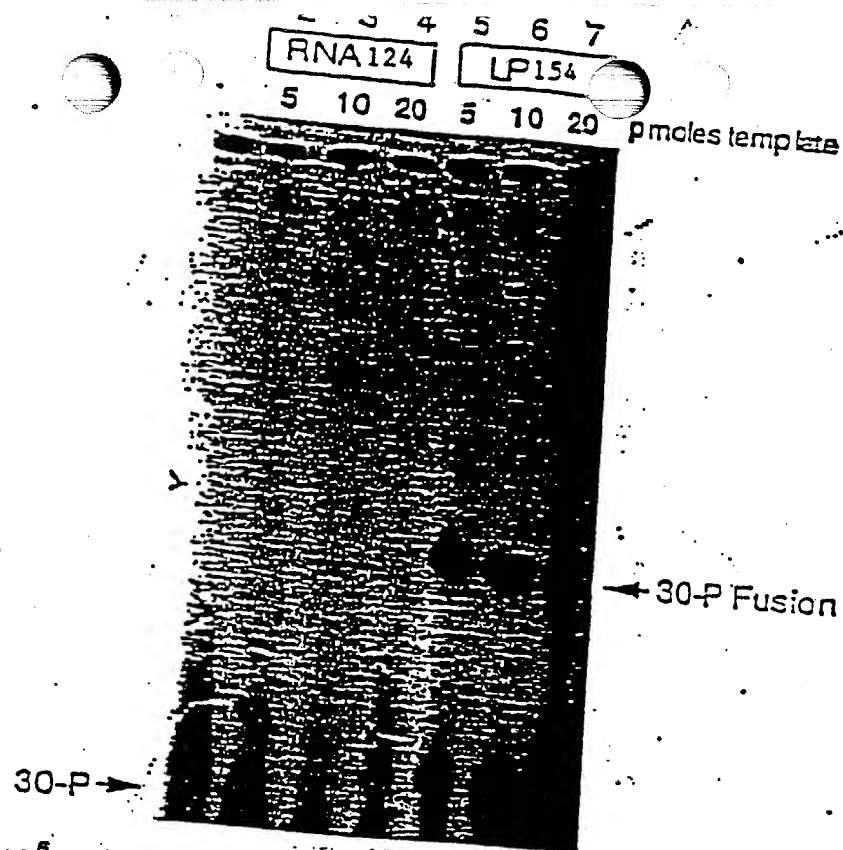


FIG. 12

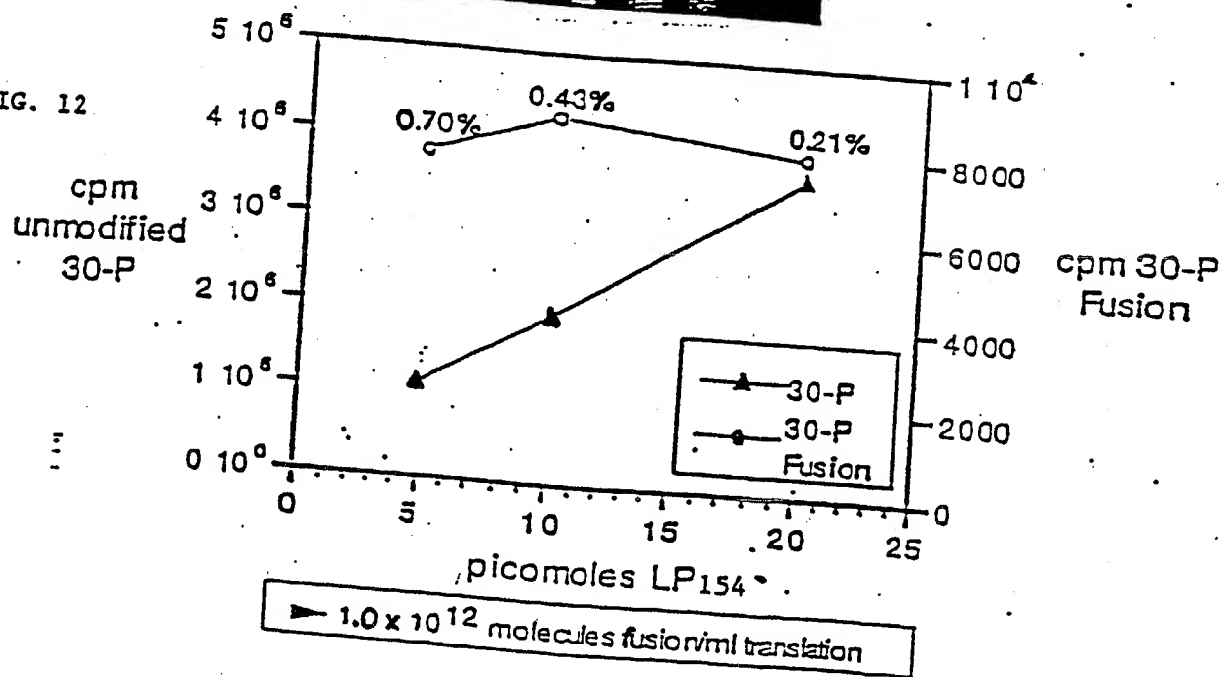


FIG. 13

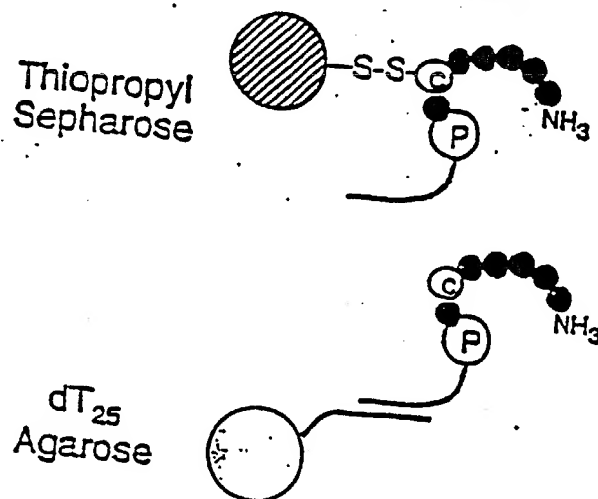
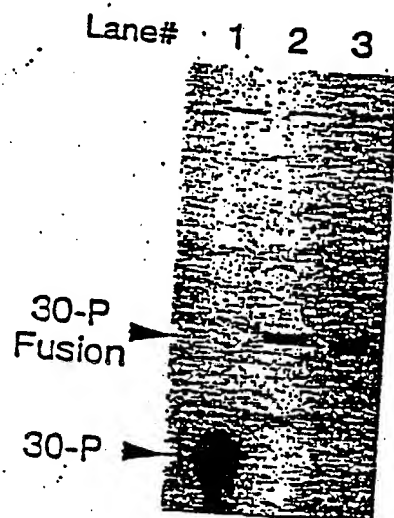


FIG. 14



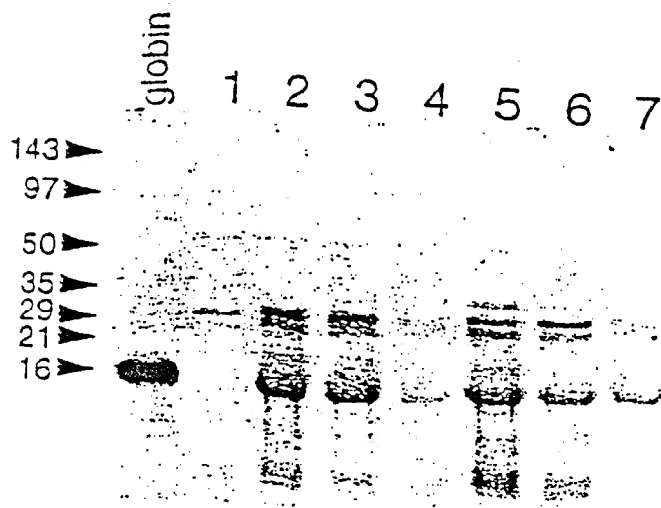


FIG. 15A

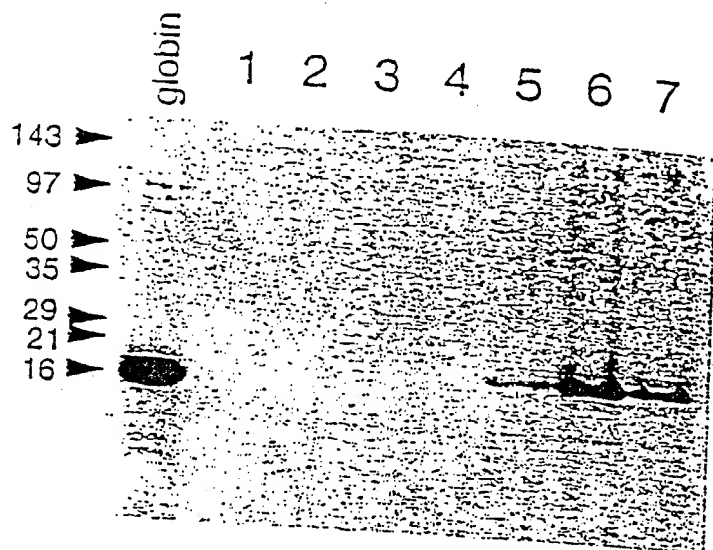


FIG. 15B

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FIG. 16 A

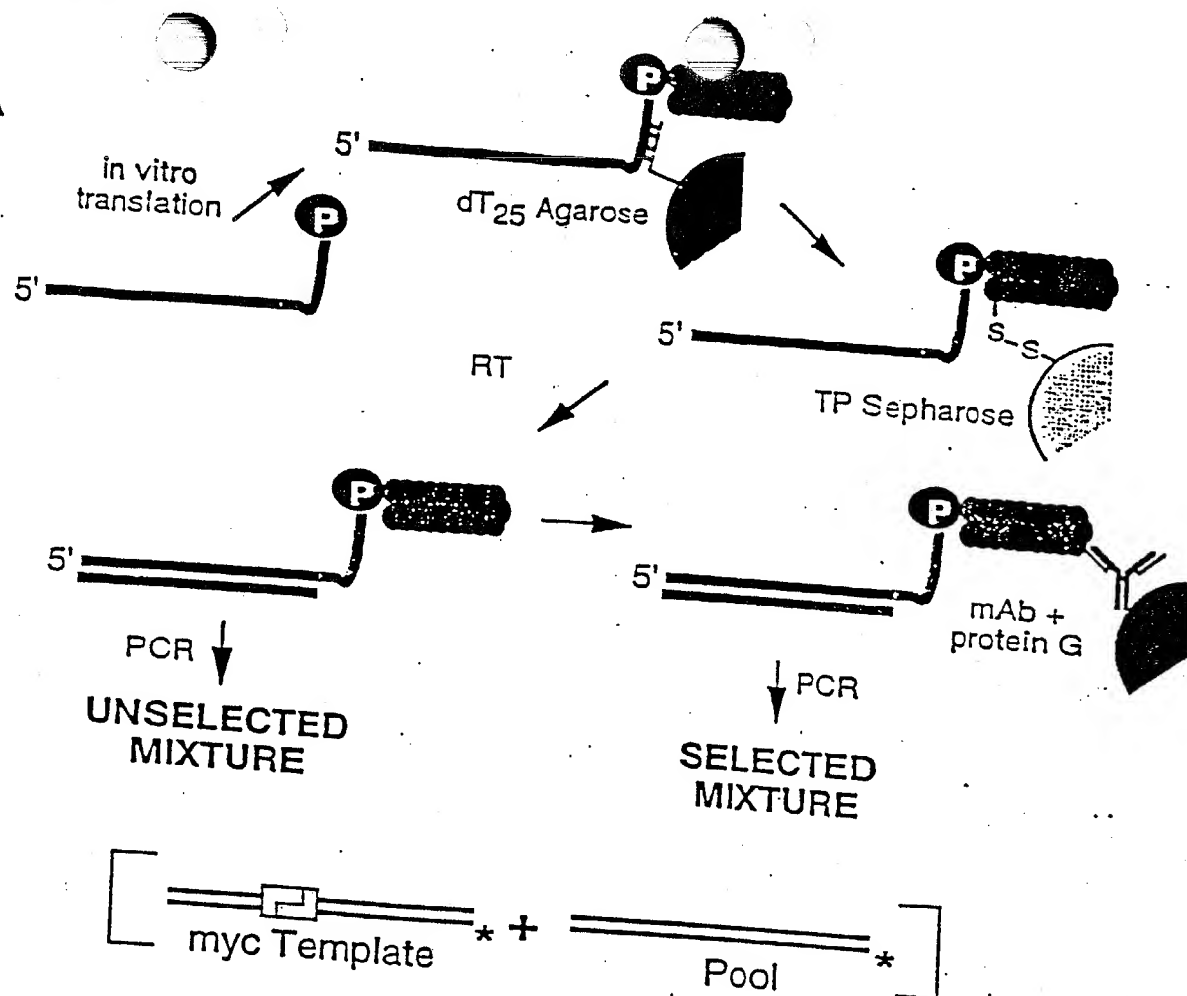


FIG. 16 B

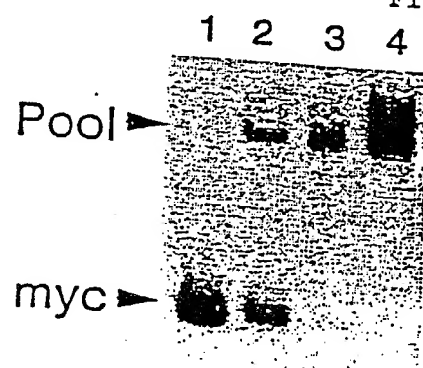
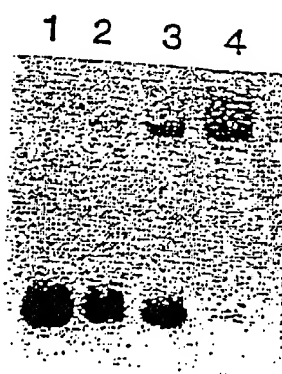


FIG. 16 C



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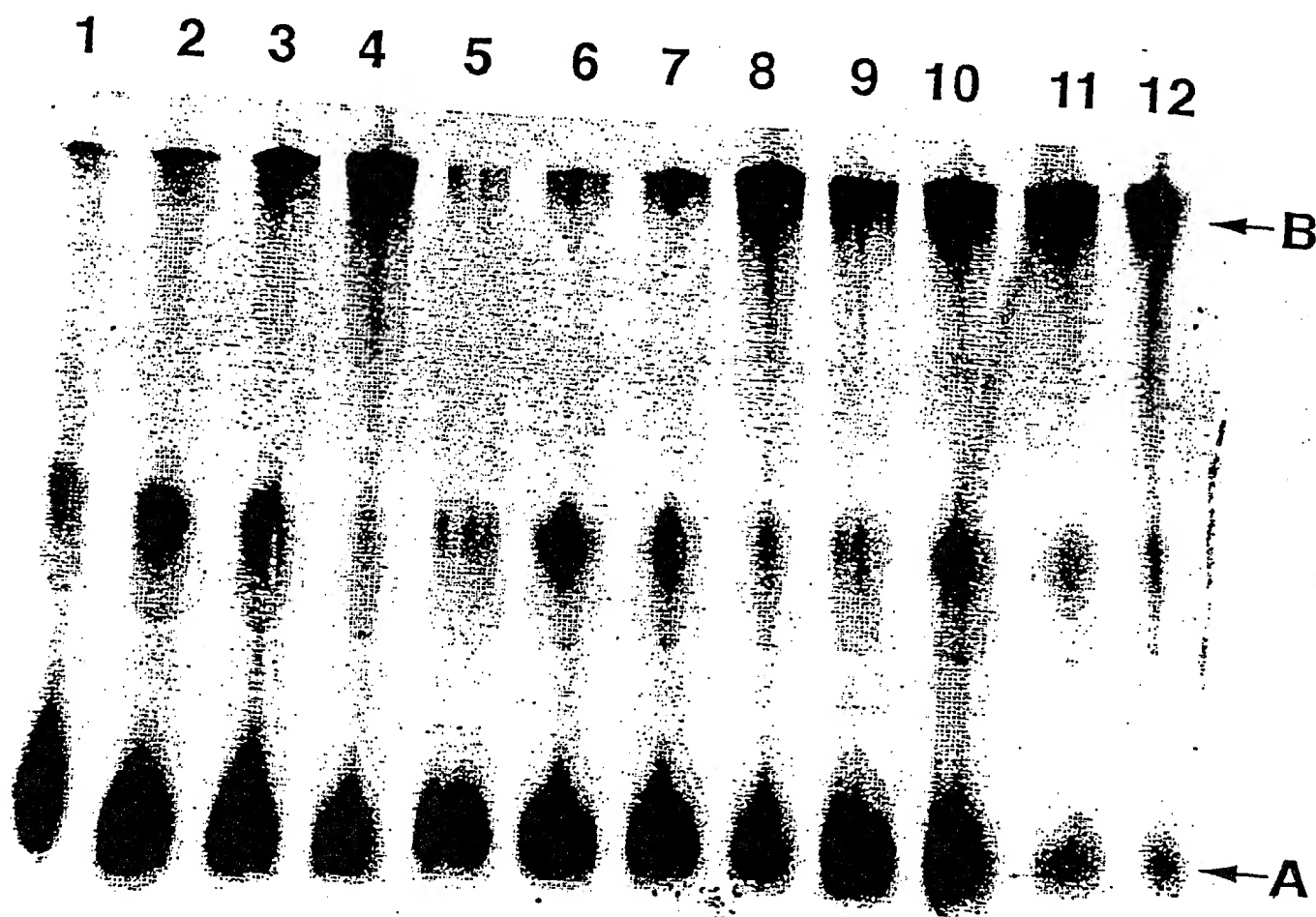


FIG. 17

09855 0650

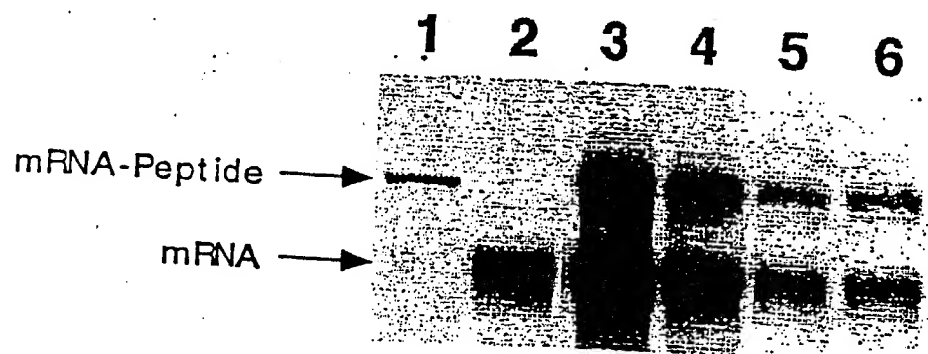


FIG. 18

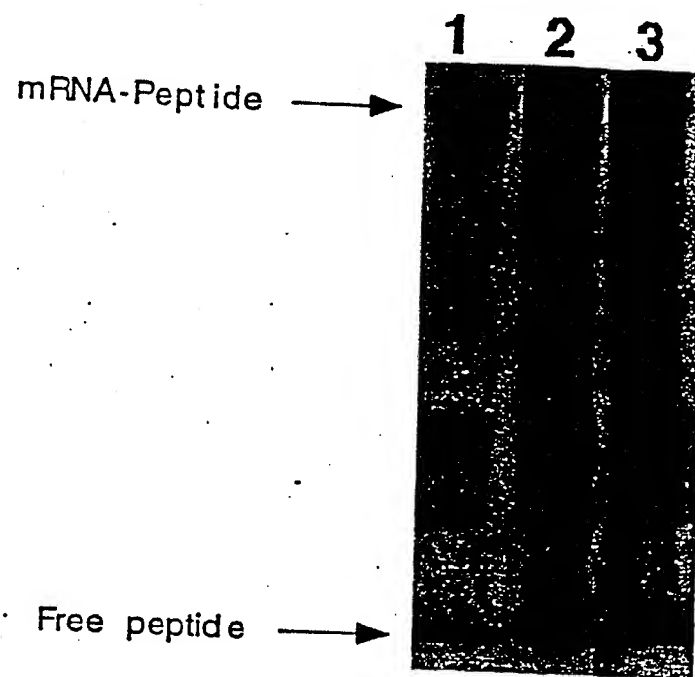


FIG. 19

FIG. 20

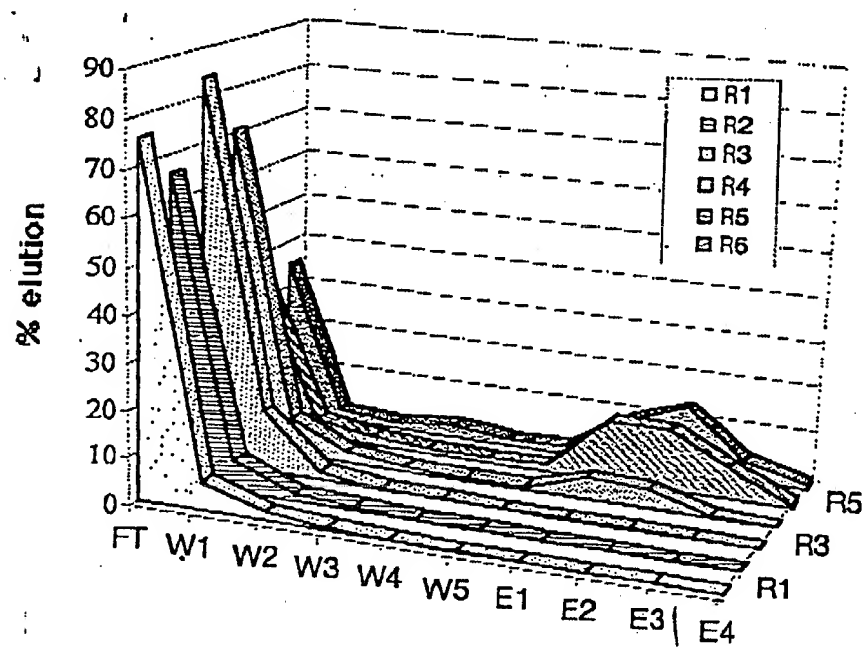


FIG. 21

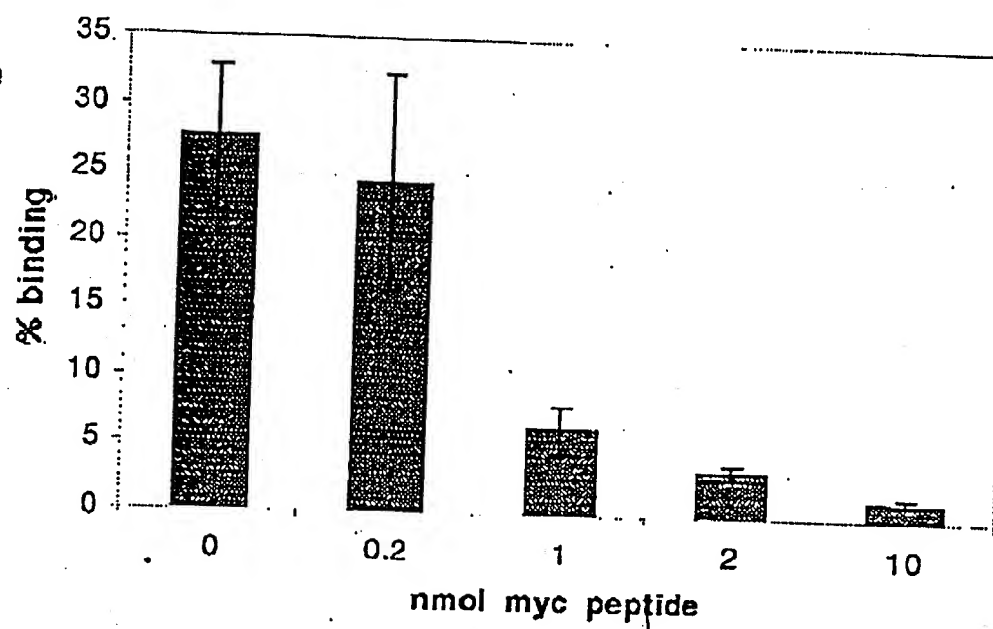


FIG. 22

c-myc epitope	E	Q	K	L	I	S	E	E	D	L
R6-51	C	A	S	V	L	S	E	R	E	C
R6-52	E	E	Y	L	V	S	E	Y	V	M
R6-53	R	Q	Y	L	I	S	E	Y	E	H
R6-55	L	Q	R	L	L	S	E	Q	M	F
R6-56	I	V	R	L	L	S	E	Y	H	M
R6-58	E	E	Y	L	L	S	E	Y	V	M
R6-60	M	Q	N	L	L	S	E	H	E	L
R6-61	T	M	D	L	L	P	E	H	Y	M
R6-63	E	Q	K	L	L	S	E	E	D	L
R6-66	D	M	M	L	L	S	E	K	E	I
R6-67	F	Q	A	L	L	A	E	E	E	I
R6-68	Q	R	V	L	L	S	E	F	W	L
consensus	X	Q/E	X	L	I	S	E	X	X	L/M

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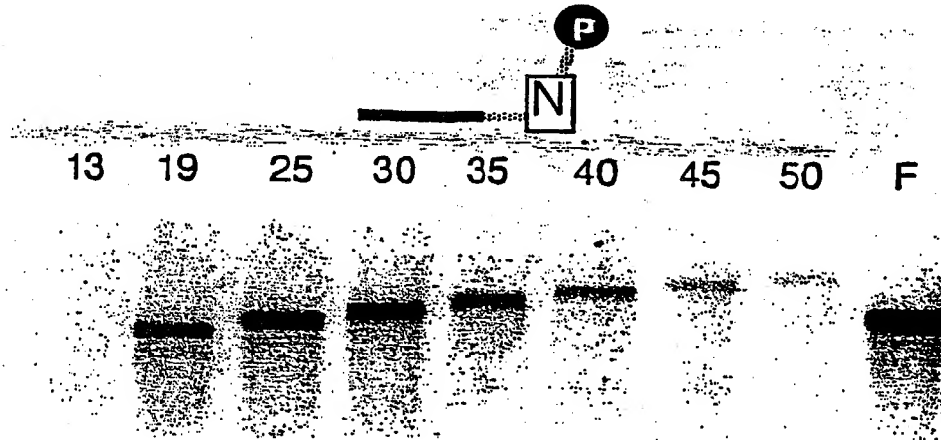


FIG. 23

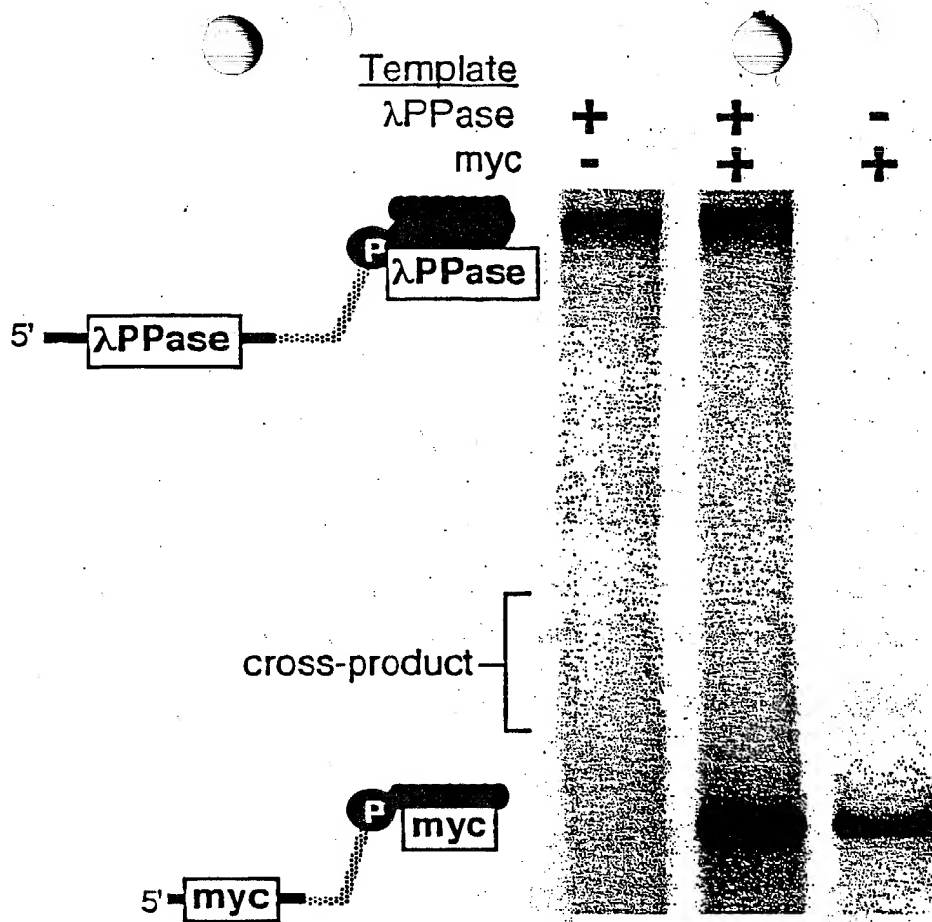


FIG. 24